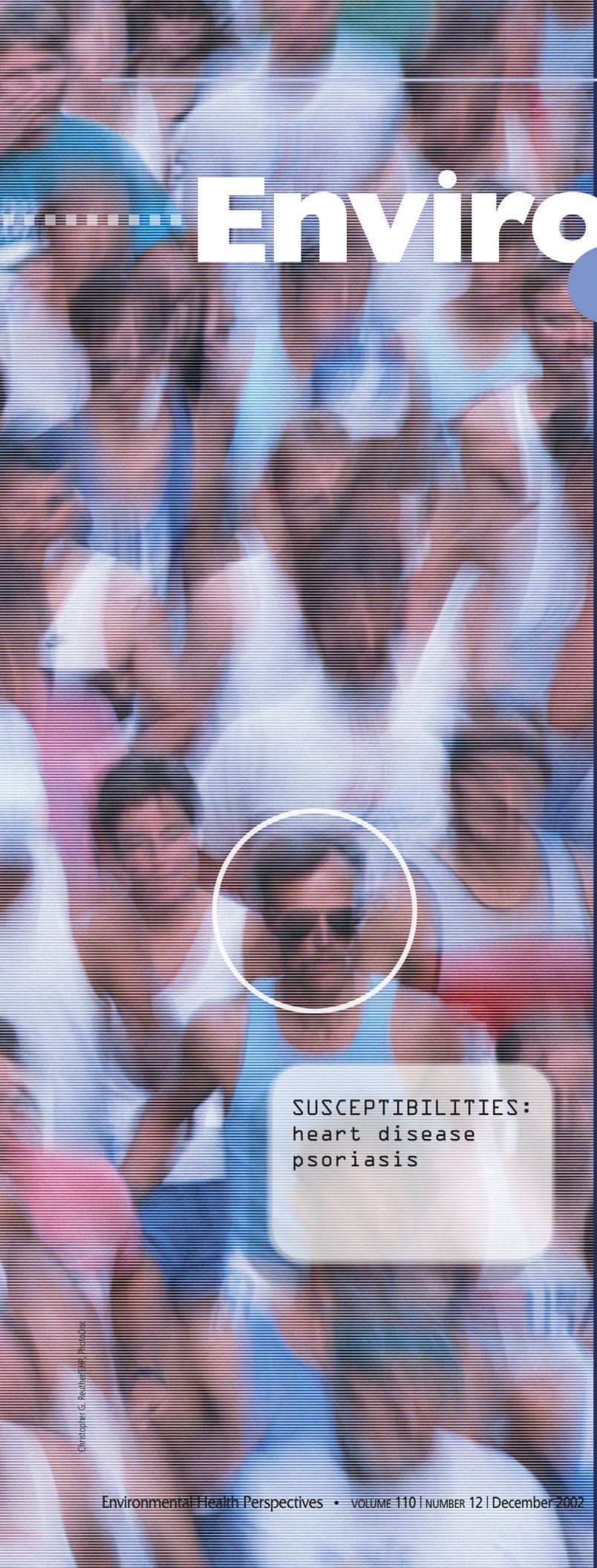
Spheres of Influence



Christopher G. Reuther/EHP; PhotoDisc

Environmental Genome Project

Focusing on Differences to Understand the Whole

As the Environmental Genome Project (EGP) marks its fifth anniversary in 2003, there is some cause for celebration. Although it is premature to expect the project to have any impact on public health and policy yet, the EGP's science and technology are advancing in step with its original goals of understanding the complex interrelationship between environmental exposure, genetic susceptibility, and human disease.

The NIEHS began the EGP in late 1997. The EGP is a multicomponent project comprising extramural and intramural research in several key areas. In addition to enhancing understanding of human genetic susceptibility to environmental exposure—how individuals differ in susceptibility to environmental agents, and how such susceptibilities change over time—other topics of interest are dose response, identification of sensitive subpopulations, and selection of appropriate test systems for human responses. EGP research is also looking at selected genes in different individuals to learn the extent and location of variations in a single DNA pair, also called single nucleotide polymorphisms, or SNPs. SNP variations are thought to be central to why people respond differently to various drugs or environmental toxicants.

The project's findings have the potential to revolutionize everything from assessing a chemical's safety, to predicting a subpopulation's adverse reactions to a new drug, to identifying earlier warning signs of much subtler effects. "The EGP will open up a whole new vista of both public health and design of medicine—molecular-designed medicines that specifically target genes that have been modified because of SNPs," says James Selkirk, deputy director of the National Center for Toxicogenomics. The center is a coordinated, multidisciplinary research program of the NIEHS that has conducted the intramural research phase of the EGP.

Since its inception, the EGP has been closely tied conceptually and through the collection of sequencing data to the Human Genome Project, which was initiated in October 1990. The EGP heavily relies on sequencing data produced from the Human Genome Project and other related sequencing projects. "[Genome mapping projects] that are going on right now are very important tools for us in environmental health," says William Farland, acting deputy assistant administrator for science in the Office of Research and Development at the U.S. Environmental Protection Agency (EPA). "The kinds of data that we are getting out of these studies will allow us to begin to identify working genes and gene products that might be at the heart of understanding public health impacts, interindividual variability, or susceptibility within populations."

Individual Susceptibility

The EGP has generated a working list of more than 550 environmentally responsive genes related to important cellular pathways such as metabolism, DNA repair, and cell cycle control that are potential targets for resequencing to identify each gene's SNPs. So far the EGP has experimentally identified 9,095 SNPs from the 123 environmentally responsive genes that have been resequenced to date, says Joan Packenham, director of the EGP's Comparative Mouse Genome Consortium. Work is under way to sequence another 200 of these genes.

The resequencing is taking place at the University of Washington, and the findings are being compiled into a central database of gene SNPs at the University of Utah Genome Center in Salt Lake City. Once researchers understand the prevalence and distribution of SNPs in those key genes, the next step is to search specific genetically susceptible subpopulations of individuals with given diseases, such as heart disease, diabetes, and genetic diseases. This could provide clues as to whether SNPs in those specific genes are related to disease formation.

Understanding what the genes actually do is key. "Once we can identify and say 'here it is, here's its variability'—that's the easy question," says George Lucier, retired associate director of the National Toxicology Program, now an NIEHS consultant and an adjunct senior toxicologist for Environmental Defense. "We're going to have to learn what a lot of these genes do and how they are related to toxic responses before we can use that information in a widespread way in risk assessment," he says. Lucier encourages more hypothesis-driven research using automated DNA sequencing. Such research would probe specific lines of queries rather than, for example, just broadly collecting resequencing data.

One EGP accomplishment of note is the creation of the Comparative Mouse Genome Consortium, which is working to develop transgenic and knockout mouse models based on variants among the environmental response genes. This consortium of academic research centers will use the mouse models to determine the functional significance of human DNA polymorphisms and analyze the function of these polymorphic variants. The consortium's research portfolio includes extramural and intramural studies to identify human DNA polymorphisms in the population and epidemiological studies to track their prevalence.

Development of technology to support such research is sorely needed, according to Daniel W. Nebert, an environmental health and genetics researcher at the University of Cincinnati. The kinks are still being worked out with new tools. For example, the advent of microarray technology means researchers can look at the expression of a variety of genes simultaneously. "This creates some advantages—you can get information more quickly—but you have to process it and sort out what's important and what's just noise," Lucier says.

"We see that polymorphism or genome technology will eventually probably merge with microarray technology to help us search for the actual phenotypes associated with susceptibility," says Raymond Tennant, director of the National Center for Toxicogenomics. "But for now, we're still searching to catalog the various variants. That's still a very formidable task."

Risk Assessment

To date, information from the EGP has been slow to trickle into the policy-making arena. "At this point the EGP has had very little influence on any risk assessment that was made. That doesn't mean it doesn't have promise. But there's really fear among all the major players about how to use this sort of information," Lucier says.

Industry leaders fear this technology could be used to call a chemical hazardous. Conversely, environmental advocates fear that industries could use this information to misidentify their chemicals as safe. Regulatory agencies, mindful of these fears, could be more hesitant in applying the technology to their own risk assessments, because they fear it lacks credibility. "That doesn't mean that it won't happen sometime in the future," Lucier says.

"[The EGP] will transform risk assessment, but we're approaching that very incrementally," Tennant agrees. Meanwhile, the volume of data is still too small to draw any conclusions, says Selkirk. First, the information needs to mature, the assays need to be validated, and researchers need to obtain a better understanding of the relevance of particular changes in gene expression and toxicity—for example, what they mean in terms of dose response and identification of sensitive subpopulations.

The genome data will help understand responses to environmental toxicants and allow for more accurate extrapolation of experimental animal responses to human populations, Farland says. Scientists will be better able to predict how a chemical will affect a certain mechanism once there is information on how the chemical acts and what pathways it disrupts.

Besides improving cross-species extrapolation, the EGP will enable "the design of epidemiological approaches that allow us to identify more subtle effects and therefore identify either susceptibilities or adverse impacts much earlier and much more quickly than we might have," Farland says. By testing individuals for various SNPs, researchers and clinicians ultimately will be able to predict whether those individuals will be vulnerable to a given chemical or whether a new drug might be harmful to them.

To expedite the process of identifying both susceptible subpopulations and the potential for adverse effects, researchers and policy makers are starting to look to hypothesis-driven research projects that will address specific questions in toxicology, particularly as it relates to structure–activity relationships and identifying key gene expression events and toxic responses. "As we then start enlarging that overall database, it will get easier and easier to ask those questions, because we'll have a better framework in which to ask them," Lucier says. "If you understand why a chemical is toxic, you can start predicting what other chemicals might be toxic, better understand dose–response relationships, better identify a sensitive subpopulation. You can use that information in risk assessment as well as other public health policy activities."

Experts believe the first applications should be chosen from classes of chemicals for which there is already sufficient background information to know what the responses mean, such as endocrine disruptors. Scientists already know many of the genes that change upon exposure to estrogens, for example. So applying chemicals with unknown estrogenic activity to that framework could show whether they act like estrogens. The technology may also be readily applied to dioxins and some metals, says Lucier.

As the fruits of the EGP's labors start appearing in the scientific literature, the EPA is beginning to bring the new data into risk assessment. Farland says current risk assessments for trichloroethylene, for instance, which are under review, actually consider these genetic factors and give some examples where either lifestyles or existing conditions or genetic factors could have an impact on the degree of hazard and risk.

"These are complex data sets, and in a lot of cases we have to filter out the important pieces of information from all of the data that are there to try to understand the implications for risk assessment," he says. "Until then, we're going to take it on a case-by-case basis. And we're going to use the process of peer involvement and peer review to make sure the scientific community is very much involved in the way that we use some of this cutting-edge type information in our work." He adds that, in the future, more cases will focus specifically on gene changes and the range of genetic variation in human populations.

Drug Development

Environmental genome data are also wending their way into the drug development and safety evaluation processes in

the pharmaceutical industries and academic and federal research centers. "The Holy Grail is to discover gene changes that are predictive of a toxic insult and that can be used in early screening of drug candidates," says I. Y. Rosenblum, group director of drug safety at the Schering Plough Research Institute. In this way, potential toxicities can be identified much earlier in the process, saving millions of dollars in development costs.

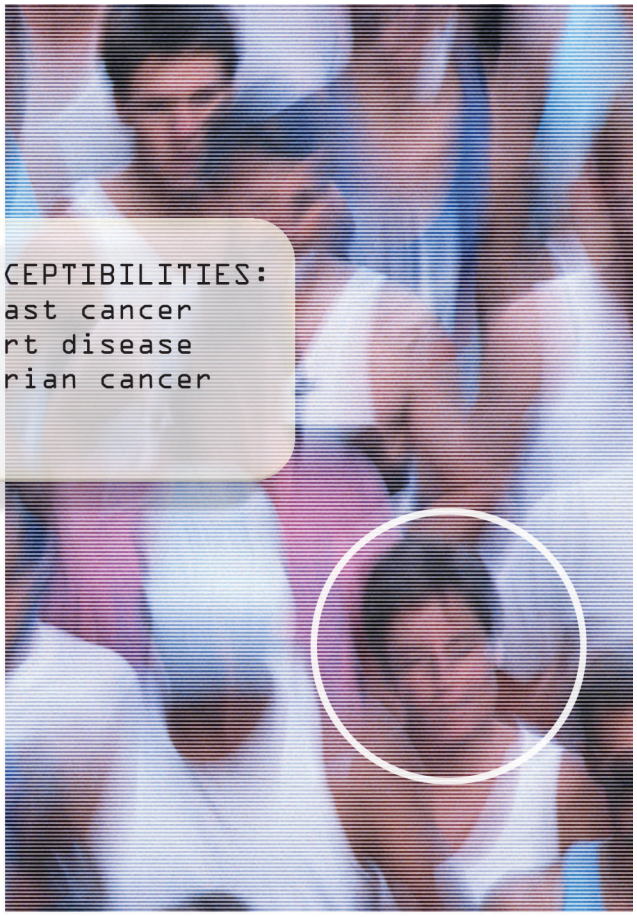
"As we understand the reasons why [some] people have adverse side effects whereas most people don't, we can determine why a drug might not be appropriate for an individual who expresses genes at a higher level or lower level than someone else," Lucier says. "No one wins when a drug that's good for ninety-eight percent of the population has a bad effect on two percent. If we could identify the two percent ahead of time, then [the drug] would be used with a great deal more confidence, especially with a good efficacious drug for the treatment of a particular disease."

Findings from the EGP and the field of toxicogenomics may also provide new drug discovery targets by using genomics methodologies to help guide selection of new candidate drugs to be put into drug development, Rosenblum says. "Many companies are aggressively building up toxicogenomic databases," he says. "There are consortiums trying to do this. The Food and Drug Administration [FDA] is involved in some of these efforts. Every major pharmaceutical company I can think of, including smaller biotech companies, are all engaged in some aspect of doing this."

Rosenblum adds that the EGP may also help better explain mechanisms of toxicity, so candidate drugs might be modified to eliminate certain toxicities. Without such knowledge, "you may be throwing the baby out with the bathwater," he says.

The FDA is starting to look into whether there are better ways to not only understand susceptible members in the population but also design better therapies. Environmental genome data are already being incorporated into evaluations in safety pharmacology, a branch of safety evaluation that integrates the best practices of pharmacology, physiology, and toxicology. For example, people with a certain type of metabolism—such as so-called slow acetylators—may be more vulnerable to a given carcinogen. Others may be protected against certain classes of chemicals, because they carry a genetic polymorphism for a detoxifying metabolic process.

The pace of progress at the level of clinical applications is also accelerating. Researchers



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are attempting to define subpopulations who will or will not respond positively to a therapeutic agent and those individuals that will more likely to suffer a toxic response. Environmental genome data have proven helpful in several cases. In one, EGP researchers at the University of Cincinnati have identified gene variants that affect the way individual asthma sufferers respond to albuterol, a drug commonly used to control acute attacks. They found a variant that can produce 8,192 combinations, 12 of which directly impact response to the medication. Researchers may now be able to identify asthma patients who will respond either well or poorly to albuterol.

Future Directions

Many challenges lay ahead for the EGP. "One leading challenge is to have the bioinformatics keep up with the data that are generated," Farland says. Statistical and computer models for analyzing gene–environment interactions are just a start. Methods to analyze macromolecular cellular components are also key. Another leading challenge is narrowing the field to identify the genes and pathways most relevant to environmental health.

As more information emerges from the EGP, the complex ethical, legal, and social issues entailed in delving so deeply into an individual's DNA are sure to become more prominent. "In terms of the policy issues, this is going to get into some discussion of ethics and the question of personal information and how much one wants to know about their genome and their susceptibility to chemicals, and who needs to know and things like that," Farland says. "These are the kinds of things ethicists are now starting to deal with, and at some point they're going to be entering into the debate in a large way."

Farland says agencies would like to see this kind of information extended into the understanding of ecologic impacts as well. "There is a fair amount of work going on that will be looking at the use of these types of data for assessing impacts on fish and invertebrates as well," he says. "So even beyond the human EGP, there will be some important data coming through on some of these other ecological species as well." Finally, says Farland, this information may be called into service as agencies get better at communicating with the public about their risks from exposure to environmental chemicals.

In the future, Packenham would like to see a large resequencing effort to include other categories of environmentally responsive genes such as those involved in signal transduction, apoptosis, oxidative stress, and drug metabolizing enzymes. To improve risk assessment, Packenham also advocates for EGP data being used to support population-based studies for the prevalence of high-risk alleles for environmentally related disease.

Much has been learned about why individuals react differently to the same environmental exposures, but with new findings come new questions about the complex interplay between environmental exposure, genetic susceptibility, and human disease—as well as how to use the information. Despite the unknowns, the EGP, with its potential for revolution, is a powerful force, and the years ahead should bring new insights into our individual differences.

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